

# Effects of vitamin D levels on asthma control and severity in pre-school children

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**Abstract. – OBJECTIVE:** Prevalence of asthma and vitamin D deficiency has been increasing and leading to significant morbidities. This study aimed to compare the vitamin D levels in the pre-school children with asthma and in healthy controls and to assess the relationship between vitamin D levels and asthma clinical parameters and control.

**PATIENTS AND METHODS:** Vitamin D [25(OH)D<sub>3</sub>] levels were measured in 102 pre-school children, aged 1-4 years with asthma and 102 healthy controls in winter. The patients with asthma were grouped according to serum vitamin D levels as sufficient, insufficient and deficient. Asthma control was classified according to the Global Initiative for Asthma (GINA) guidelines and the Test for Respiratory and Asthma Control in Kids (TRACK) in 1-4 years-old children.

**RESULTS:** Serum vitamin D levels were 22.64 (9.96) ng/ml in the asthma group and 32.11 (14.74) ng/ml in the control group ( $p = 0.001$ ). Total number of exacerbations during the previous year were significantly lower in the vitamin D sufficient group, compared to the deficient and insufficient groups ( $p = 0.03$ ). Frequency of patients with controlled asthma was higher in the sufficient group compared to the deficient and insufficient groups ( $p = 0.001$  and  $p = 0.001$ , respectively). There was a positive correlation between serum vitamin D levels and asthma control.

**CONCLUSIONS:** The frequency of vitamin D deficiency and insufficiency was higher in children with asthma, compared to the controls. Therefore, we suggest that lower levels of vitamin D are associated with poor asthma control and increased asthma severity.

*Key Words:*

Asthma, Vitamin D, Pre-school children, Asthma control, Test for respiratory and asthma control in kid.

## Introduction

Asthma is among the common chronic inflammatory diseases in childhood and the first

symptoms are observed in childhood in half of the cases<sup>1</sup>. Incidence and cost of asthma in both developed and developing countries have been increasing. Asthma is one of the most significant morbidities of childhood due to the school absenteeism, emergency room visits and hospitalization<sup>1,2</sup>. Currently, the prevalence of vitamin D deficiency has been increasing parallel with asthma<sup>3</sup>. Natural vitamin D production (approximately 80%) is identified by the ultraviolet (UV) component of sunlight through cutaneous synthesis<sup>4-6</sup>. The adoption of a Western-type lifestyle, altered eating habits, and spending less time doing outdoor activities are thought to play a role in the increased prevalence of vitamin D deficiency<sup>7-8</sup>. Vitamin D has been suggested to contribute to asthma pathogenesis by improving immune function, exhibiting anti-inflammatory effects, reducing the steroid resistance, increasing the effects of glucocorticoids, slowing the cell cycle down, and reducing remodeling<sup>3,4,9,10</sup>.

Many studies have reported a possible relationship between asthma and vitamin D deficiency; however, evidence is still controversial. Some cross-sectional studies<sup>7,11-14</sup> have shown that serum vitamin D levels in asthmatic children are lower than in healthy children. Other studies have not observed such a significant relationship<sup>15-19</sup>, whereas a limited number of studies have shown that increased serum vitamin D levels may increase the risk of asthma<sup>20,21</sup>. In addition, vitamin D levels and asthma severity, exacerbation, emergency room visits, and duration of hospitalization may vary according to the studies in asthma patients<sup>4,11,19,22,23</sup>.

There are a limited number of studies which investigated the relationship between asthma control and vitamin D levels. The data obtained

from these studies mostly include school-age children, adolescent and adult origin. In addition, there are no studies examining the relationship between vitamin D and asthma control in preschool children.

The present study aimed to compare vitamin D levels in pre-school children with asthma and in healthy controls and to assess the relationship between vitamin D levels and asthma clinical parameters, as well as the effects on asthma control.

## **Patients and Methods**

### ***Study Population***

One hundred two children with mild-to-moderate asthma according to the Global Initiative for Asthma (GINA) who were admitted to the Pediatric Immunology and Allergy outpatient clinic at Eskisehir State Hospital Between December 2014 and February 2015 were enrolled. The control group consisted of 102 healthy children admitted to the general pediatric outpatient clinic aged 1-4 years. The diagnosis of asthma was made by clinical characteristics of cough and wheezing during exercise, laughing, or crying in the absence of a respiratory infection varies over time and which is reversible with bronchodilator administration, at least 3 documented episodes of wheezing as suggested by GINA guidelines<sup>1</sup>. Patients and control group who had a history of consumption of any supplements of vitamin D or drugs that modulate serum vitamin D levels, such as systemic corticosteroids, anticonvulsants, and those who had acute chronic diseases were excluded. In addition, the presence of allergies was also an exclusion criterion.

### ***Study Design and Ethics Committee Approval***

This case-control study was approved by the Ethics Committee of Eskisehir State Hospital. The study was performed in accordance with the principles of the Helsinki Declaration. Written informed consent was obtained from each child and his/her parents.

### ***Data Collection***

Age, gender, height, weight, parental allergic diseases (asthma, allergic rhinoconjunctivitis and atopic dermatitis), and sunlight exposure (at least 10 minutes between 10 and 15 hours) were recorded<sup>24</sup>.

The Asthma Symptom Score (ASS) consisted of respiration rate, retractions, dyspnea and wheezing, each of which was rated 0-3; 0 being the least, 3 being the most severe<sup>25</sup>. Emergency Department visits, number of in-hospital stays, number of exacerbations within the last year were recorded.

According to the GINA assessment of asthma severity, the pediatric allergy specialist assessed asthma severity in consideration of daytime and nighttime symptoms, the number of exacerbations, and values as mild and moderate asthma<sup>1</sup>. According to the GINA assessment of asthma control in children 5 years and younger, the patients assessed asthma control in consideration daytime and nighttime symptoms, any activity limitation due to asthma and reliever medication needed more than once a week (in the past 4 weeks) values as "well controlled, partly controlled and uncontrolled"<sup>1</sup>.

The TRACK questionnaire suggested in measurement of asthma control in children under 4 years old by GINA was completed by parents or caregivers. The TRACK questionnaire completed by the parents or caregivers is a test that consists of five items that measure asthma control. Each item reflects the clinical points lowest as "0" and as highest "20" including 5 questions<sup>26</sup>.

The questionnaire consists of the frequency of respiratory problems per week such as wheezing, coughing, or shortness of breath occurring during the past four weeks, the frequency of sleep disturbances at night per week, the frequency of interference with playing the games, going to school, or participating in normal activities for her/his age, the frequency of given rescue treatment within the past three months (relaxing medication) and during the last 12 months, and the frequency of using systemic corticosteroids due to respiratory problems uncontrollable by other drugs. The sum of these items provided the total score, ranging from 0 to 100. The higher scores indicated better disease control (controlled > 80), whereas a score of less than 80 (uncontrolled < 80) suggested potential respiratory problems. The reliability and validity of TRACK was demonstrated by Yilmaz et al<sup>27</sup>.

Peripheral venous blood samples were obtained to measure serum Vitamin D, parathormon (PTH), calcium (Ca), phosphorus (P), alkaline phosphatase (ALP), immunoglobulin E (IgE) levels, and peripheral blood eosinophil

was counted on the same day. Ca, P and ALP were measured by spectrophotometrically in the biochemistry laboratory (AU 5800 Clinical Chemistry Analyzer, Beckman Coulter Inc., USA).

### **Vitamin D and PTH Measurement**

25 (OH) D3 was measured by ECLIA (electrochemiluminescence immunoassay; Cobas E601 Immunoassay System, Roche Diagnostics, Indianapolis, IN, USA), parathormon (PTH) CLIA (chemiluminescence immunoassay; Architect I2000SR Immunoassay System, Abbott Diagnostics, Lake Forest, IL, USA) in the hormon laboratory.

The values for vitamin D levels > 30 ng/ml were considered as sufficient, those between 20 and 30 ng/ml as insufficient, and lower than 20 ng/ml as deficient<sup>28</sup>.

### **Total Eosinophil Counts**

Eosinophil counts were recorded from a complete blood count device (Cell Dyne 3700 Analyzer, Abbott Diagnostics, Lake Forest, IL, USA).

### **Total IgE Measurement**

Two milliliters of venous blood samples were collected into standard biochemical tubes. These samples were studied by chemiluminescence immunometric method (IMMULITE 2000 Immunoassay System, Siemens Healthcare, Malvern, PA, USA) in the biochemistry laboratory.

### **Statistical Analysis**

The statistical analysis was performed using the SPSS 15.0 software program (SPSS Inc., Chicago, IL, USA). The Kolmogorov-Smirnov test was used to analyze the normally distributed data. The Student's *t*-test and ANOVA were used to compare normally distributed data between independent groups. Quantitative data are expressed as mean  $\pm$  standard deviation. The Mann-Whitney U-test and Kruskal-Wallis test were used for independent groups with abnormal distribution. Quantitative data are expressed as median (interquartile range). Pearson's chi-square (linear by linear correlation) was used to compare non-parametric variables (with a preset probability of  $p < 0.05$ ). The correlation of vitamin D levels and clinical variables were analyzed by the Spearman's correlation analysis (with a preset probability of  $p < 0.05$ ). A  $p$  value < 0.05 was considered statistically significant.

## **Results**

### **Sociodemographic Characteristics of Asthmatic Children and Healthy Controls**

The median (interquartile range) ages of the asthmatic and healthy control children were 3.0 (2.5-4.0) years and 3.0 (2.0-4.0) years, respectively ( $p = 0.49$ ). Total of 55.9% of the patients and 51.0% of the control subjects were males (Table I). Age, height, weight, and sunlight exposure were similar between the patients and

**Table I.** Sociodemographic characteristics and laboratory findings of the children in the study.

	Asthma group (n = 102)	Control group (n = 102)	$p^*$
Age	3.0 (2.5-4.0)	3.0 (2.0-4.0)	0.49
Gender (male)	57 (55.9)	52 (51.0)	0.48\$
Height (cm)	95.0 (88.0-100.0)	96.0 (88.0-101.3)	0.57
Weight (kg)	14.0 (12.4-15.0)	14.0 (13.0-16.0)	0.11
Parental atopy presence	56 (54.9)	41 (40.2)	0.04\$
Sunlight exposure (10 minute >)	16 (15.7)	24 (19.6)	0.16\$
Vitamin D levels (ng/ml)	22.64 (9.96)	32.11 (14.74)	0.001 <sup>†</sup>
Vitamin D status			0.001\$
Deficient (< 20 ng/mL)	47 (46.1)	25 (24.5)	
Insufficient (20-30 ng/mL)	31 (30.4)	24 (23.5)	
Sufficient (> 30 ng/mL)	24 (23.5)	53 (52)	
Parat Hormone (ng/ml)	37.05 (25.82-45.68)	31.15 (22.88-39.75)	0.02
Ca	9.9 (0.4)	10.3 (0.5)	0.001 <sup>†</sup>
P	4.9 (0.6)	5.5 (0.7)	0.001 <sup>†</sup>
ALP	210.0 (182.3-256.3)	189.5 (164.5-237.3)	0.02
Eo	192.0 (93.5-328.5)	180.0 (88.7-300.0)	0.78
IgE	34.1 (16.5-107.2)	15.5 (6.8-39.3)	0.001

\*Mann-Whitney U-test (expressed in median-interquartile range); <sup>†</sup>Student *t*-test. \$Pearson's Chi-Square (n-%).

control groups. Parental atopy was 56 (54.9%) in the asthma group and 41 (40.2%) in the control group ( $p = 0.04$ ) (Table I).

### Laboratory Findings of Asthmatic Children and Healthy Controls

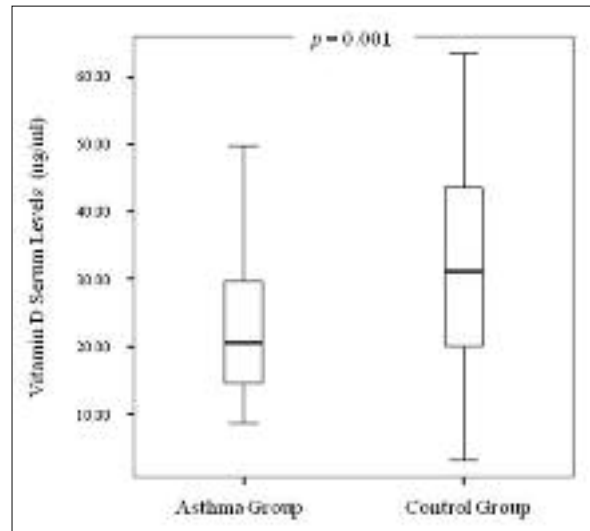
Vitamin D levels were 22.64 (9.96) ng/ml in the patient group and 32.11 (14.74) ng/ml in the control group, indicating statistical significance ( $p = 0.001$ ) (Figure 1) (Table I). When the asthma patients were divided into groups according to the levels of vitamin D, rates of deficiency was 47 (46.1%), insufficiency was 31 (30.4%), sufficiency 24 (23.5%) in the asthma group while deficiency 25 (24.5%), insufficiency 24 (23.5%), sufficiency 53 (52%) in the control group. Vitamin D status different between the groups ( $p = 0.001$ ) (Table I).

Serum PTH, Ca, P, and ALP levels were significant different between the asthma and control groups ( $p = 0.02$ ;  $p = 0.001$ ;  $p = 0.001$  and  $p = 0.02$  respectively) (Table I).

None of the children in the patient and control groups had active rickets.

### Serum IgE and Eosinophil Counts

The asthma patients exhibited significantly higher IgE levels compared to the control group median (interquartile range) 34.1 (16.5-107.2) IU/ml and 15.5 (6.8-39.3) IU/ml, respectively; ( $p = 0.001$ ) (Table I). The eosinophil counts did not differ significantly between the asthma and control groups ( $p = 0.78$ ) (Table I).



**Figure 1.** The box plot of serum vitamin D levels in children with asthma and control groups.

### Sociodemographic Characteristics of Children with Asthma According to Vitamin D Levels

The median (interquartile range) ages of the deficient, insufficient, and sufficient groups were 3.5 (2.5-4.0) years, 3.0 (2.0-4.0) years, and 2.0 (3.3-4.0) years, respectively ( $p = 0.35$ ). Males represented 55.3% of the deficient group, 58.1% of the insufficient group, and 54.2% of the sufficient group ( $p = 0.95$ ) (Table II).

The presence of height, weight, and parental atopy did not significantly differ between the

**Table II.** Relationship between serum levels of vitamin D and sociodemographic and laboratory findings of the patient group.

	Deficient ( $< 20$ ng/ml) ( $n = 47$ )	Insufficient ( $20-30$ g/ml) ( $n = 31$ )	Sufficient ( $> 30$ ng/ml) ( $n = 24$ )	$p^*$
Age	3.5 (2.5-4.0)	3.0 (2.0-4.0)	2.0 (3.3-4.0)	0.35
Gender (male)	26 (55.3)	18 (58.1)	13 (54.2)	0.95 <sup>s</sup>
Height (cm)	14.5 (13.5-15.5)	14.0 (11.5-15.0)	13.8 (11.0-14.4)	0.35
Weight (kg)	95.6 (8.2)	92.0 (9.1)	91.5 (9.6)	0.13 <sup>†</sup>
Parental atopy presence	27 (57.4)	16 (51.6)	13 (54.2)	0.88 <sup>s</sup>
Sunlight exposure (10 minute >)	3 (6.4)	4 (12.9)	9 (37.5)	0.003 <sup>s</sup>
Vitamin D levels (ng/ml)	14.03 (2.79)	24.65 (3.22)	36.91 (6.10)	0.001 <sup>†</sup>
Parathormone (ng/ml)	42.6 (35.2-49.0)	34.4 (25.0-45.3)	27.1 (20.3-38.2)	0.002
Ca	9.9 (0.4)	9.9 (0.5)	10.0 (0.5)	0.5 <sup>†</sup>
P	5.0 (0.7)	5.0 (0.6)	5.0 (0.6)	0.47 <sup>†</sup>
ALP	212.0 (186-255)	213 (177-257)	202.5 (166-259.5)	0.79
Eo	185 (101-321)	198 (95-403)	201.5 (51-273.5)	0.8
IgE	46.4 (17-124)	26.8 (16.5-97.4)	28.9 (16.5-97.1)	0.48

<sup>s</sup>Pearson's Chi-Square [% (n)]; \*Kruskal Wallis Test-Chi Square; <sup>†</sup>ANOVA.

groups ( $p > 0.05$  for all) (Table II). The patient ratio receiving adequate sunlight exposure was 6.4%, 12.9 and 37.5% in the deficient, insufficient, and sufficient groups, respectively, suggesting a statistical significance ( $p = 0.003$ ) (Table II).

#### **Laboratory Finding According to the Vitamin D Status of Children with Asthma**

Vitamin D levels in children with asthma were 14.03 (2.79) ng/ml in deficient group, 24.65 (3.22) ng/ml in the insufficient group and 36.91 (6.10) ng/ml in the sufficient group ( $p = 0.001$ ), while parathormon levels were 42.6 (35.2-49.0) ng/ml in the deficient group, 34.4 (25.0-45.3) ng/ml in the insufficient group, and 27.1 (20.3-38.2) ng/ml in the sufficient group ( $p = 0.002$ ) (Table II). Serum Ca, P and ALP levels did not differ significantly between deficient, insufficient and sufficient groups ( $p > 0.05$  for all) (Table II).

Serum IgE levels and eosinophil counts did not differ significantly between the deficient, insufficient, and sufficient groups ( $p = 0.48$  and  $p = 0.8$ , respectively) (Table II).

#### **Clinical Characteristics According to the Vitamin D Status in Children with Asthma**

The median (interquartile range) age of initial asthma diagnosis in the asthma groups were not

significant ( $p = 0.07$ ). The median (interquartile range) Asthma Symptoms Scores did not differ significantly between the asthma groups ( $p = 0.28$ ) (Table III).

The median (interquartile range) number of asthma exacerbations within the last year was higher in the deficient and insufficient group, compared to the sufficient group 3 (2-4), 3 (1-4) and 2 (1-2,8), respectively ( $p = 0.03$ ) (Table III). No statistically significant difference was observed in terms of the number of emergency service visits and the number of hospitalizations within the last year among the groups ( $p = 0.15$  and  $p = 0.51$ , respectively) (Table III).

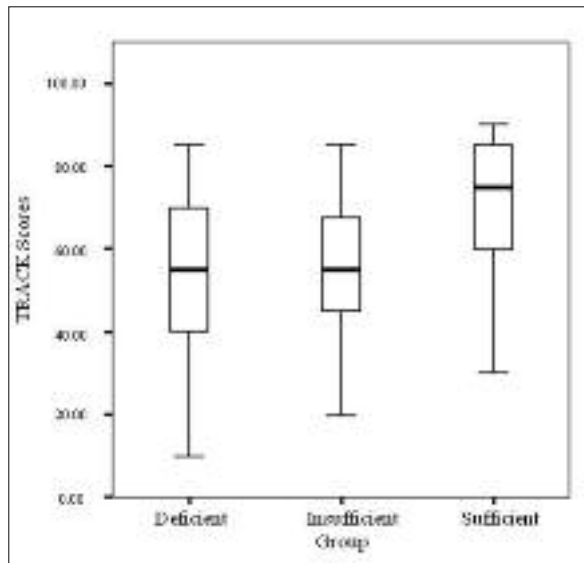
Asthma severity showed a statistically significant difference among the vitamin D groups. Mild asthma rate was 36.2%, 41.9% and 75% in deficient, insufficient and sufficient groups, respectively whereas moderate asthma rate was 63.8%, 58.1% and 25% in deficient, insufficient and sufficient groups ( $p = 0.006$ ) (Table III).

The mean TRACK score was 52.2 (19.8), 55.5 (19.8), and 70.2 (16.9) in the deficient, insufficient, and sufficient groups, respectively ( $p = 0.001$ ) (Figure 2) (Table III). According to the TRACK scoring, 10.6% asthma was controlled in the deficient group, 16.1% in the insufficient group, and 45.8% in the sufficient group. Uncontrolled asthma (TRACK scores  $< 80$ ) was 89.4%, 83.9%, and 54.2% in the deficient, insufficient, and sufficient groups; therefore the difference among the groups was significant ( $p = 0.001$ )

**Table III.** Relationship between serum levels of vitamin D and sociodemographic and laboratory findings of the patient group.

	Deficient ( $< 20$ ng/ml) (n = 47)	Insufficient (20-30 ng/ml) (n = 31)	Sufficient ( $> 30$ ng/ml) (n = 24)	$p^*$
Duration of disease (months)	18 (6-24)	10 (4-18)	13.5 (9.8-18)	0.07
Asthma Symptoms Scores	2 (1-3)	2 (1-3)	1 (1-3)	0.28
Asthma morbidity (last year)				
Number of exacerbation	3 (2-4)	3 (1-4)	2 (1-2,8)	0.03
Number of emergency service visits	3 (2-4)	2 (3-5)	2 (2.5-3)	0.15
Number of hospitalizasyon	0 (0-1)	0 (0-1)	0 (0-1)	0.51
Asthma severity				0.006 <sup>§</sup>
Mild	17 (36.2)	13 (41.9)	18 (75.0)	
Moderate	30 (63.8)	18 (58.1)	6 (25.0)	
Asthma Control (GINA)				0.001 <sup>§</sup>
Well control	4 (8.5)	3 (9.7)	17 (70.8)	
Partial control	18 (38.3)	16 (51.6)	3 (12.5)	
Uncontroll	25 (53.2)	12 (38.7)	4 8 (16.7)	
TRACK Scores	52.2 (19.8)	55.5 (19.8)	70.2 (16.9)	0.001 <sup>†</sup>
Control	5 (10.6)	5 (16.1)	11 (45.8)	0.001 <sup>§</sup>
Uncontrol	42 (89.4)	26 (83.9)	13 (54.2)	

<sup>§</sup>Pearson's Chi-Square [% (n)]; \*Kruskal Wallis Test-Chi Square; <sup>†</sup>ANOVA.



**Figure 2.** The box plot of TRACK Scores in children grouped according to the Vitamin D levels.

(Table III). When patients were classified according to TRACK score those with controlled asthma had higher vitamin D levels compared to patients with uncontrolled asthma group (30.04 ng/ml vs. 20.95 ng/ml,  $p = 0.001$ )

According to GINA, well asthma control was 70.8% in the sufficient group, 8.5% in the deficient group, and 9.7% in the insufficient group and the difference between the groups was statistically significant ( $p = 0.001$ ) (Table III). Vitamin D levels were 32.09 ng/ml, 20.19 ng/ml, and 19.32 ng/ml in well, partially and uncontrolled asthma groups, respectively. There were statistically significant differences between the groups ( $p = 0.001$ ) (Figure 3).

#### **Correlation Between Serum Vitamin D Levels and Clinical Variables**

Vitamin D levels were correlated with the duration of sunlight exposure ( $r = 0.25$ ;  $p = 0.01$ ), number of exacerbations in the last year ( $r = -0.27$ ,  $p = 0.006$ ). Asthma severity, asthma control, and TRACK score were correlated with serum vitamin D level ( $r = -0.34$ ,  $r = 0.46$  and  $r = 0.29$  respectively, and  $p = 0.001$ ,  $p = 0.001$ , and  $p = 0.004$ , respectively) (Figure 3) (Table IV).

Age, gender, parental atopy presence, IgE levels and eosinophil count were not correlated with serum vitamin D level ( $r = -0.80$ ,  $r = 0.05$ ,  $r = 0.02$ ,  $r = -0.15$  and  $r = -0.07$  respectively,  $p > 0.05$  for all).

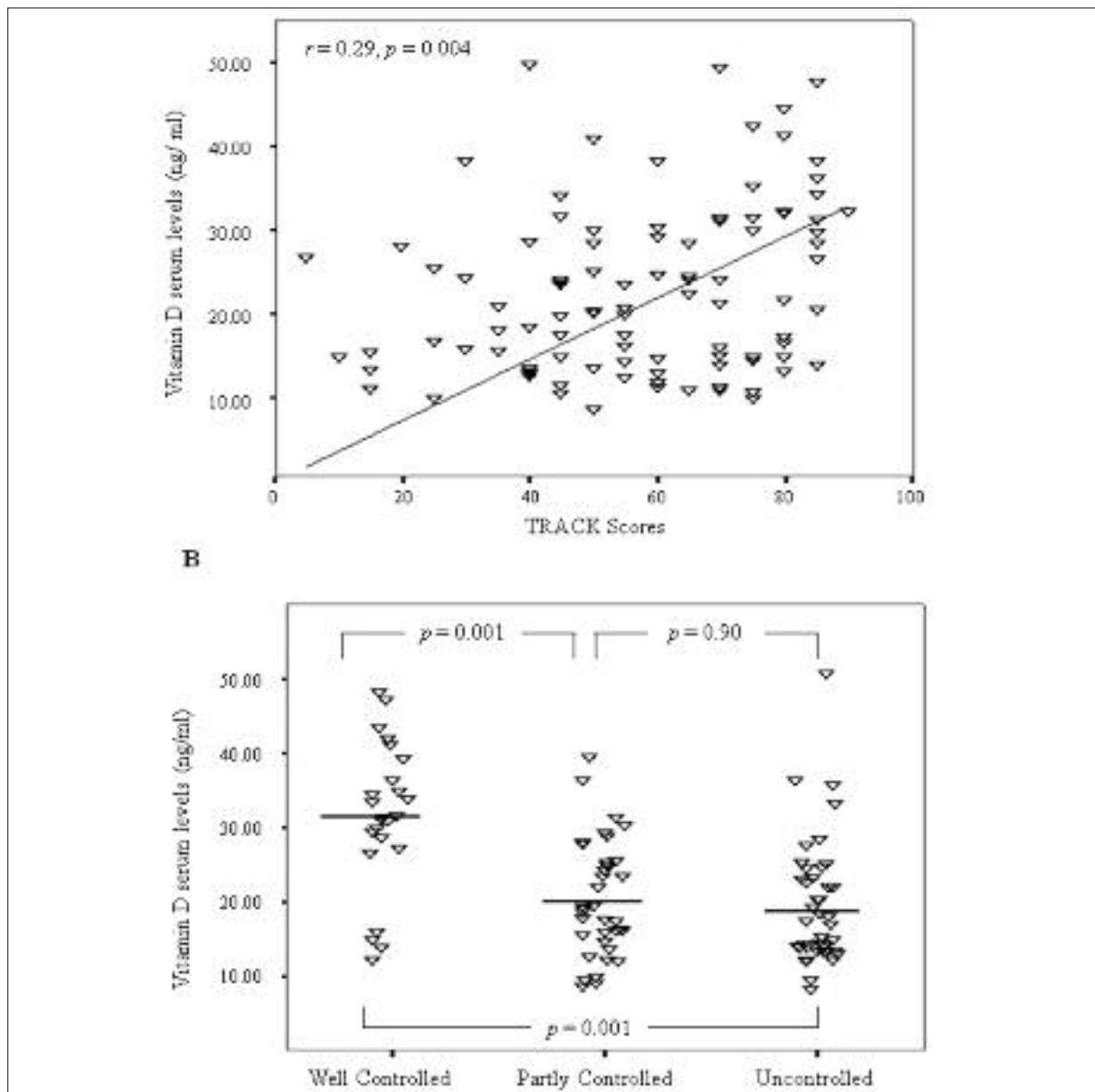
Similarly, duration of disease, number of emergency service visits, and the number of hospitalization within the last year and ASS was not correlated with serum vitamin D level ( $r = -0.07$ ,  $r = -0.14$ ,  $r = -0.11$ ,  $r = -0.15$ ,  $r = 0.17$  and  $r = -0.11$  respectively,  $p > 0.05$  for all) (Table IV).

## **Discussion**

In the present study, vitamin D levels in preschool children with asthma were lower than the healthy controls. When asthma patients were classified as deficient ( $< 20$  ng/ml), insufficient (20-30 ng/ml), and sufficient ( $> 30$  ng/ml) according to their vitamin D levels, asthma control rates according to GINA and TRACK were statistically higher in the sufficient group, compared to the deficient and insufficient group. In addition, there was a positive correlation between level of asthma control and vitamin D levels. The number of exacerbations within the last year was lower in the sufficient group compared to the other groups. The percentage of sunlight exposure was significantly lower in deficient and insufficient groups than the sufficient group.

The development of asthma is associated with the large number of immunological markers. Asthma prevalence and vitamin D deficiency rates have been increasing worldwide. An increasing body of evidence supports the pleiotropic effects of vitamin D on various chronic disorders including those associated with immune regulatory function. This is associated with a number of childhood diseases such as type 1 diabetes mellitus, celiac disease, and asthma<sup>3</sup>. The reduction in vitamin D production on the skin through short-term exposure to sunlight, and inadequate access to food and supplements lead to vitamin D deficiency in children in particular. This supports the idea that there is a decrease in the time spent in open areas due to the Western lifestyle. For instance, people in the United States have been estimated to spend approximately 93% of their time indoors<sup>7,8</sup>.

The serum levels of 25(OH) D<sub>3</sub> are measured as a marker of vitamin D status due to its long half-life (~15 days). There are still no studies showing the effects of vitamin D in initiating asthma. The results of high 25 (OH) D<sub>3</sub> levels related to reducing the incidence of asthma is controversial. The birth cohort study in Australia concluded that low 25 (OH) D<sub>3</sub> levels at the age of 6 have been shown to be predictive of later de-



**Figure 3.** The relationship existing between vitamin D serum levels and asthma control **A**, Scatter plot shows the relationship existing between vitamin D serum levels and TRACK scores. **B**, Distribution of vitamin D serum levels in children grouped according to the “GINA assessment of asthma control in children 5 years and younger”. Horizontal lines show mean values.

veloping atopy or asthma phenotypes in 14-years-old boys<sup>29</sup>. Similarly, Van Oeffelen et al<sup>20</sup> showed that elevated vitamin D levels at the age of 4 decrease the asthma risk at the age of 8. On the contrary, Tolpanen et al<sup>21</sup> claimed that high 25 (OH) D<sub>3</sub> levels at the age of 10 increased the asthma incidence and wheezing risk in subsequent years.

Vitamin D levels have been shown to be statistically significantly lower in asthmatic chil-

dren compared to the healthy children in studies of Mediterranean countries such as Italy, Cyprus, and Turkey, as well as Middle Eastern countries such as Iran and Qatar<sup>7,15,25,30,31</sup>. As a result of the current study, vitamin D levels in asthmatic children were determined to be lower compared to the control group, similar to other studies. The reason for the low levels of vitamin D in asthmatic children compared to the control group is spending more time in indoor areas due to exac-

**Table IV.** Univariate analysis of associations between serum Vitamin D levels and clinical variables\*.

Variable	Correlation*	p*
Age	-0.80	0.42
Gender	0.05	0.60
Parental atopy presence	0.02	0.84
Sunlight exposure (10 minute >)	0.25	0.01
IgE	-0.15	0.14
Eo	-0.07	0.48
Duration of disease, months	-0.07	0.51
Asthma morbidity (last year)		
Number of exacerbation	-0.27	0.006
Number of emergency service visits	-0.14	0.17
Number of hospitalization	-0.11	0.27
Asthma Symptoms Scores	-0.11	0.27
Asthma severity	-0.34	0.001
Asthma Control (GINA)	0.46	0.001
TRACK Scores	0.29	0.004

\*Correlation was determined with the Spearman rank correlation coefficient.

exacerbation and protecting the infections triggering this and as a result they were exposed to less sunlight. However, Dogru et al<sup>19</sup> showed that there was no significant difference between asthmatic children and healthy subjects in Turkey. This difference may arise from the difference in weather during which the two studies were performed. Moreover, it may be also caused by the lower age compared to other groups of patients constituting our study group.

In a cross-sectional study<sup>32</sup> conducted on children at the age of 5-11 in Italy, vitamin D deficiency was determined in 53.5% of the children with asthma and insufficiency in 37.3%, while vitamin D sufficiency was detected in 9.4% of children. Deficient, insufficient, and sufficient rates were 43.3%, 47.5%, and 9.2% respectively in a study conducted with children at the ages of 3-8.5 in our country. In another study conducted with children at the ages of 2-14, the rates were 67%, 20%, and 9.5%. The deficiency rate was 46.1% in our study, which was similar to other studies<sup>25</sup>, while insufficiency and sufficient was 30.4% and 23.5%, respectively. Higher vitamin D sufficiency rate in our study compared to the other studies may be explained by the older ages of children in other studies and seasonal differences. The study from 2001-2006, the National Health and Nutrition Examination Survey<sup>33</sup>, which showed that the vitamin D deficiency in children aged 1-5 was lower than those who are 6-11, supports our results.

Evidence suggests that there are multiple biological effects of vitamin D in the pathogenesis of respiratory diseases such as asthma and allergic diseases, as well as bone metabolism<sup>34</sup>. Vitamin D deficiency can affect the lung development and functions<sup>35</sup>. *In vivo* and *in vitro* animal model studies demonstrate that vitamin D has an important role in modulation of normal lung development; thus, vitamin D deficiency may impair lung development<sup>36</sup>. Furthermore, vitamin D has been shown to have various effects in the innate and adaptive immune systems. Airway epithelium, which is an element of the innate immune system against the external area, contains a high level of enzyme converting 25(OH)D<sub>3</sub> to an active form of 25(OH)D<sub>3</sub>. This active form of vitamin D has a local effect on infection responses. Therefore, it is effective in reducing inflammation due to infections<sup>9</sup>. Vitamin D affects the immune system by increasing the expression of cathelicidin, which is an important antimicrobial peptide against respiratory pathogens<sup>10,14</sup>. Observational studies<sup>8,37,38</sup> suggest that vitamin D deficiency in children and adults can contribute to the incidence of wheezing and increases the risk of respiratory infections that cause asthma exacerbations. In a randomized placebo controlled study conducted by Urashima et al<sup>39</sup>, influenza A infection was seen in Japanese school-age children taking 1200 IU of vitamin D 3 per day. Interestingly, less exacerbation was observed in subgroup of asthmatic children receiving vitamin D supplements compared to the control group. Vitamin D also effects the adaptive immune system through Th<sub>1</sub>, Th<sub>2</sub>, and regulatory T cells (T reg)<sup>9</sup>. Vitamin D promotes induction of T reg cells providing the controlling of inhibitor cytokine secretion (IL-10 and TGF- $\beta$ ) and CD4<sup>+</sup> T lymphocytes (Th<sub>1</sub>, Th<sub>2</sub>). Vitamin D was shown to inhibit the Th17 cytokine production related to asthma severity and low steroid response. Vitamin D can also increase glucocorticoid responsiveness in steroid resistant asthma<sup>3,5,10</sup>. Vitamin D deficiency decreases corticosteroid responsiveness, contributing to asthma attacks<sup>4</sup>. By these mechanisms, vitamin D may also have a therapeutic role in reducing asthma attacks<sup>4</sup>.

In the general population, despite controversial evidence on the association of vitamin D with asthma thus far, there is an inverse relationship between sufficient levels of vitamin D and asthma medication use, and asthma severity indicators such as exacerbation and hospitalization in children with asthma<sup>4,11,19,22,23</sup>. In the present study,



number of asthma exacerbations within the last year was higher in the deficient and insufficient groups, compared to the sufficient group, while no difference was observed in the number of hospitalizations. Our results are consistent with the results of Dogruoz et al<sup>19</sup>. In Childhood Asthma Management Program (CAMP) study conducted with 1024 mild-moderate persistent asthmatic children, low vitamin D levels were retrospectively associated with increased odds of hospitalization during the previous year, and prospectively with severe asthma exacerbations over the following four years<sup>9</sup>. Our study group involved mild to moderate asthmatic patients and a negative correlation was observed between the numbers of the previous year's exacerbation and vitamin D levels, similar to the CAMP study, while there was no relationship between hospitalization numbers<sup>11</sup>. The results of the study that lacked a correlation between the number of hospitalizations and vitamin D levels showed similar results as the study of Allen et al<sup>40</sup>.

There was no relationship between vitamin D levels and atopy markers IgE and eosinophil numbers in patients with asthma in our study. The results of current study are similar to the results of Alyasin et al<sup>40</sup>, although not with the results of Brehm et al<sup>11</sup> study conducted in Costa Rica.

Currently, the primary aim is to control asthma with asthma guidelines. However, there is no standard definition of adequate asthma control. There are a limited number of studies assessing the relationship between vitamin D and asthma control in children. In a recent cross-sectional observational study of 75 Italian children with asthma, aged 5 to 11 years, Chinellato et al<sup>31</sup> documented significant and positive correlations between serum 25 (OH) D<sub>3</sub> levels and improved spirometric measurements of airway obstruction and a common measure of asthma control (GINA and the Childhood Asthma Control Test). Lower vitamin D levels were associated with poor asthma control and lower lung function. According to GINA<sup>25</sup>, significant differences in the level of asthma control and levels of vitamin D has been shown in a study conducted with asthmatic children aged 2-14 in Turkey. Gupta et al<sup>23</sup> conducted a study that involved steroid resistance and moderate asthmatics and found a positive relationship between 25 (OH) D<sub>3</sub> levels and ACT, whereby higher serum 25 (OH) D<sub>3</sub> was associated with better asthma control. Serum vitamin D was significantly lower in elderly patients with uncontrolled asthma compared to the ones with

controlled disease<sup>41</sup>. In a placebo controlled study of Lewis et al<sup>42</sup>, high-dose vitamin D treatment was initiated for deficiency and sufficiency in asthmatic patients between the ages of 6-17 for one year. There was no significant difference in measurements of vitamin D, spirometric values, and ACT levels in the placebo and treated groups at the end of the year. However, ACT data obtained from all visits were pooled, and a positive correlation was found between serum 25 (OH) D<sub>3</sub> levels and ACT scores (Pearson's  $r = 0.25$ ,  $p = 0.05$ ), suggesting an effect of vitamin D in asthma management. These results showed that asthma control was better in the sufficient group compared to the other groups according to the classification of TRACK and GINA measuring asthma control in children under 4 years old. A positive correlation was found between vitamin D levels and asthma control, consistent with the aforementioned studies. However, Alyasin et al<sup>40</sup> did not identify any significant relationship between vitamin D levels and asthma control according to GINA in children aged 6-18.

However, the present study had some limitations. The sample size was relatively small ( $n = 204$ ). A larger sample size would have increased the statistical power to detect associations. We did not consider the effect of vitamin D consumed by diet in study group.

## Conclusions

The rate of vitamin D insufficiency and deficiency were higher in asthmatic children compared to the controls. We also found a strong inverse relationship between circulating levels of vitamin D and asthma control parameters. We also showed that greater exacerbation, increased asthma severity, and reduced asthma control was associated with vitamin D deficiency and insufficiency.

## Conflict of Interest

There are no conflicts of interest for any of the authors. Authors of this manuscript state that the submitted manuscript does not overlap with another manuscript.

## References

- 1) 2014 GINA REPORT, Global strategy for asthma management and prevention. Updated December 2011. Available from: <http://www.ginasthma.org/GINA-Report,-Global-Strategy-for-Asthma-Management-and-Prevention>

- 2) ASHER I, PEARCE N. Global burden of asthma among children. *Int J Tuberc Lung Dis* 2014; 18: 1269-1278.
- 3) RAJABBIK MH, LOTFI T, ALKHALED L, FARES M, EL-HAJJ FULEIHAN G, MROUEH S, AKL EA. Association between low vitamin D levels and the diagnosis of asthma in children: a systematic review of cohort studies. *Allergy Asthma Clin Immunol* 2014; 11: 31.
- 4) SEARING DA, ZHANG Y, MURPHY JR, HAUKE PJ, GOLEVA E, LEUNG DY. Decreased serum vitamin D levels in children with asthma are associated with increased corticosteroid use. *J Allergy Clin Immunol* 2010; 25: 995-1000.
- 5) MAGNUS MC, STENE LC, HÅBERG SE, NAFSTAD P, STIGUM H, LONDON SJ, NYSTAD W. Prospective study of maternal mid-pregnancy 25-hydroxyvitamin D level and early childhood respiratory disorders. *Paediatr Perinat Epidemiol* 2013; 27: 532-541.
- 6) LITONJUA AA, WEISS ST. Is vitamin D deficiency to blame for the asthma epidemic? *J. Allergy Clin Immunol* 2007; 120: 1031-1035.
- 7) BENER A, EHlayel MS, Tulic MK, Hamid Q. Vitamin D deficiency as a strong predictor of asthma in children. *Int Arch Allergy Immunol* 2011; 57: 168-175.
- 8) LITONJUA AA. Childhood asthma may be a consequence of vitamin D deficiency. *Curr Opin Allergy Clin Immunol* 2009; 9: 202-207.
- 9) BREHM JM, SCHUEMANN B, FUHLBRIGGE AL, HOLLIS BW, STRUNK RC, ZEIGER RS, WEISS ST, LITONJUA AA. Serum vitamin D levels and severe asthma exacerbations in the Childhood Asthma Management Program study. *J Allergy Clin Immunol* 2010; 26: 52-58.
- 10) DELLA GIUSTINA A, LANDI M, BELLINI F, BOSONI M, FERRANTE G, ONORARI M, TRAVAGLINI A, PINGITORE G, PASALACQUA G, TRIPODI S. Vitamin D, allergies and asthma: focus on pediatric patients. *World Allergy Organ J* 2014; 7: 27.
- 11) BREHM JM, ACOSTA-PÉREZ E, KLEI L, ROEDER K, BARMADA M, BOUTAOUI N, FORNO E, KELLY R, PAUL K, SYLVIA J, LITONJUA AA, CABANA M, ALVAREZ M, COLÓN-SEMI-DEY A, CANINO G, CELEDÓN JC. Vitamin D insufficiency and severe asthma exacerbations in Puerto Rican children. *Am J Respir Crit Care Med* 2012; 186: 140-146.
- 12) EHlayel M, BENER A, SABBABH A. Is high prevalence of vitamin D deficiency evidence for asthma and allergy risks? *Eur Ann Allergy Clin Immunol* 2011; 43: 81- 88.
- 13) FREISHTAT RJ, IOBAL SF, PILLAI DK, KLEIN CJ, RYAN LM, BENTON AS, TEACH SJ. High prevalence of vitamin D deficiency among inner-city African American youth with asthma in Washington, DC. *J Pediatr* 2010; 156: 948-952.
- 14) GINDE AA, MANSBACH JM, CAMARGO CA Jr. Association between serum 25-hydroxyvitamin D level and upper respiratory tract infection in the Third National Health and Nutrition Examination Survey. *Arch Intern Med* 2009; 169: 384-390.
- 15) CHINELLATO I, PIAZZA M, SANDRI M, PERONI DG, CARDINALE F, PIACENTINI GL, BONER AL. Serum vitamin D levels and exercise-induced bronchoconstriction in children with asthma. *Eur Respir J* 2011; 37: 1366-1370.
- 16) MAALMI H, BERRAÏES A, TANGOUR E, AMMAR J, ABID H, HAMZAOU K, HAMZAOU A. The impact of vitamin D deficiency on immune T cells in asthmatic children: a case – control study. *J Asthma Allergy* 2012; 5: 11-19.
- 17) MENON J, MARANDA L, NWOSU BU. Serum 25-hydroxyvitamin D levels do not correlate with asthma severity in a case-controlled study of children and adolescents. *J Pediatr Endocrinol Metab* 2012; 25: 673-679.
- 18) GERGEN PJ, TEACH SJ, MITCHELL HE, FREISHTAT RF, CALATRONI A, MATSUI E, KATTAN M, BLOOMBERG GR, LIU AH, KERCSMAR C, O'CONNOR G, PONGRACIC J, RIVERA-SANCHEZ Y, MORGAN WJ, SORKNESS CA, BINKLEY N, BUSSE W. Lack of a relation between serum 25-hydroxyvitamin D concentrations and asthma in adolescents. *Am J Clin Nutr* 2013; 97: 1228-1234.
- 19) DOGRU M, KIRMIZIBEKMEZ H, YESILTEPE MUTLU RG, AKTAS A, OZTURKMEN S. Clinical effects of vitamin D in children with asthma. *Int Arch Allergy Immunol* 2014; 164: 319-325.
- 20) VAN OEFFELEN AA, BEKKERS MB, SMIT HA, KERKHOF M, KOPPELMAN GH, HAVEMAN-NIES A, VAN DER A DL, JANSEN EH, WJUGA AH. Serum micronutrient concentrations and childhood asthma: the PIAMA birth cohort study. *Pediatr Allergy Immunol* 2011; 22: 784-793.
- 21) TOLPPANEN A, SAYERS A, GRANELL R, FRASER WD, HENDERSON J, LAWLOR DA. Prospective Association of 25-Hydroxyvitamin D3 and D2 with Childhood Lung Function, Asthma, Wheezing, and Flexural Dermatitis. *Epidemiology* 2013; 24: 310- 319.
- 22) BREHM JM, CELEDÓN JC, SOTO-QUIROS ME, AVILA L, HUNNINGHAKE GM, FORNO E, LASKEY D, SYLVIA JS, HOLLIS BW, WEISS ST, LITONJUA AA. Serum vitamin D levels and markers of severity of childhood asthma in Costa Rica. *Am J Respir Crit Care Med* 2009; 179: 765-771.
- 23) GUPTA A, SJOUKES A, RICHARDS D, BANYA W, HAWRYLOWICZ C, BUSH A, SAGLANI S. Relationship between serum vitamin D, disease severity, and airway remodeling in children with asthma. *Am J Respir Crit Care Med* 2011; 184: 1342-1349.
- 24) UYSALOL M, MUTLU LC, SARACOGLU GV, KARASU E, GUZEL S, KAYAOGULU S, UZEL. Childhood asthma and vitamin D deficiency in Turkey: is there cause and effect relationship between them? *Ital J Pediatr* 2013; 39: 78.
- 25) LIU LL, GALLAHER MM, DAVIS RL, RUTTER CM, LEWIS TC, MARCUSE EK. Use of a respiratory clinical score among different providers. *Pediatr Pulmonol* 2004; 37: 243-248.
- 26) MURPHY KR, ZEIGER RS, KOSINSKI M, CHIPPS B, MELLON M, SCHATZ M, LAMPL K, HANLON JT, RAMACHANDRAN S.

- Test for respiratory and asthma control in kids (TRACK): a caregiver-completed questionnaire for preschool-aged children. *J Allergy Clin Immunol* 2009; 123: 833-839.
- 27) YILMAZ O, TURKELI A SAHIN S, YUKSEL H. Predictive Value of the TRACK Questionnaire as a Measure of Asthma Control in Preschool Aged Children. *Allergy Asthma Immunol Res* 2014; 6: 357-361.
  - 28) HOLICK MF. Vitamin D deficiency. *N Engl J Med* 2007; 357: 266-281.
  - 29) HOLLAMS EM, HART PH, HOLT BJ, SERRALHA M, PARSONS F, DE KLERK NH, ZHANG G, SLY PD, HOLT PG. Vitamin D and atopy and asthma phenotypes in children: a longitudinal cohort study. *Eur Respir J* 2011; 38: 1320-1327.
  - 30) KOLOKOTRONI O, PAPADOPOULOU A, MIDDLETON N, KOUTA C, RAFTOPOULOS V, NICOLAIDOU P, YIALLOUROS PK. Vitamin D levels and status amongst asthmatic and non-asthmatic adolescents in Cyprus: a comparative cross-sectional study. *BMC Public Health* 2015; 15: 48.
  - 31) HATAMI G, GHASEMI K, MOTAMED N, FIROOZBAKHT S, MOVAHED A, FARROKHI S. Relationship between Vitamin D and Childhood Asthma: A Case – Control Study. *Iranian Journal of Pediatrics* 2014; 24: 710-714.
  - 32) CHINELLATO I, PIAZZA M, SANDRI M, PERONI D, PIACENTINI G, BONER AL. Vitamin D serum levels and markers of asthma control in Italian children. *J. Pediatr.* 2011; 158: 437-441.
  - 33) MANSBACH JM, GINDE AA, CAMARGO CA JR. Serum 25-hydroxyvitamin D levels among US children aged 1 to 11 years: do children need more vitamin D? *Pediatrics* 2009; 124: 1404-1410.
  - 34) BOZZETTO S, CARRARO S, GIORDANO G, BONER A, BARALDI E. Asthma, allergy and respiratory infections: the vitamin D hypothesis. *Allergy* 2011; 67: 10-17.
  - 35) ZOSKY GR, BERRY LJ, ELLIOT JG, JAMES AL, GORMAN S, HART PH. Vitamin D deficiency causes deficits in lung function and alters lung structure. *Am J Respir Crit Care Med* 2011; 183: 1336-1343.
  - 36) FOONG RE, ZOSKY GR. Vitamin D deficiency and the lung: disease initiator or disease modifier? *Nutrients* 2013; 5: 2880-2900.
  - 37) GINDE AA, MANSBACH JM, CAMARGO CA JR. Association between serum 25-hydroxyvitamin D level and upper respiratory tract infection in the Third National Health and Nutrition Examination Survey. *Arch Intern Med* 2009; 169: 384-390.
  - 38) GINDE AA, MANSBACH JM, CAMARGO CA JR. Vitamin D, respiratory infections, and asthma. *Curr Allergy Asthma Rep* 2009; 9: 81-87.
  - 39) URASHIMA M, SEGAWA T, OKAZAKI M, KURIHARA M, WADA Y, IDA H. Randomized trial of vitamin D supplementation to prevent seasonal influenza A in school-children. *Am J Clin Nutr* 2010; 91: 1255-1260.
  - 40) ALYASIN S, MOMEN T, KASHEF S, ALIPOUR A, AMIN R. The relationship between serum 25 hydroxy vitamin d levels and asthma in children. *Allergy Asthma Immunol Res* 2011; 3: 251-255.
  - 41) KORN S, HÜBNER M, JUNG M, BLETNER M, BUHL R. Severe and uncontrolled adult asthma is associated with vitamin D insufficiency and deficiency. *Respir Res* 2013; 22: 14-25.
  - 42) LEWIS E, FERNANDEZ C, NELLA A, HOPP R, GALLAGHER JC, CASALE TB. Relationship of 25-hydroxyvitamin D and asthma control in children. *Ann Allergy Asthma Immunol* 2012; 108: 281-282.